

## Mycological Research News<sup>1</sup>

This issue of Mycological Research News features: In this issue; The coronomycin producer: a case of mistaken identity?; Long-range self-organization in *Dictyostelium*; and Resistance variation in natural pathosystems.

Thirteen research papers are included in this part, seven concerning the phylogenetic relationships of plant pathogens: a new invasive *Phytophthora* on trees, variation and mating systems in *P. ramorum*, the status of the *Microdochium nivale* varieties, the position of *Pseudocercospora cladosporioides* on olives, *Verticillium s. lat.*, an ancestral powdery mildew, and a consideration of *Sawadaea* spp. along with their maple hosts. Other papers report on parthenosomes in corticioid fungi, collembola grazing, the cultivation of *Cordyceps unilateralis*, genetic diversity in *Entomophaga maimaiga*, the gold content of macromycetes, and a fossil *Aspergillus* species from Tertiary amber.

The following new scientific names are introduced: *Caespitotheca* gen. nov.; *Aspergillus collembolorum*, and *Phytophthora kernoviae* spp. nov.; and *C. forestalis* (syn. *Uncinula forestalis*), and *Microdochium majus* (syn. *Fusarium nivale* var. *majus*) combs. nov.

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### IN THIS ISSUE

The first two papers concern *Phytophthora* species which are important pathogens of trees and ornamentals. The new species *P. kernoviae* described from south-west England causes bleeding lesions on beech and stem necrosis of rhododendron (pp. 853–859). In addition, new information on the morphology and mating behaviour of the severe pathogen *P. ramorum*, causal agent of sudden oak death in North America but also present on a range of trees and ornamental shrubs in Europe, is presented based on studies of strains from Europe and North America. All but one European isolate belonged to mating type A1, while A1 and A2 mating types were both well-represented in North America; some morphological differences were also observed (pp. 860–871).

*Microdochium nivale* has long been recognized as having two varieties, var. *majus* and var. *nivale*. Now, phylogenetic analysis of sequences of the EF-1 alpha gene, leave no doubt that the varieties merit recognition at species rank, a decision supported by differences in the sizes and septation of the conidia and also in pathogenicity (pp. 872–880). Molecular phylogenetic studies using three genes show that *Pseudocercospora cladosporioides*, causal agent of Cercospora leaf spot of

olives, is correctly placed in that genus; further, the populations are genetically rather homogenous, at least in Spain, suggesting spraying may be efficacious in controlling the disease (pp. 881–888). Molecular and immunochemical studies on 12 species placed in *Verticillium s. lat.* support the revised classifications into different genera, and crossed electrophoreses clarify the relationships of *Verticillium s. str.* species and also support the recognition of *V. longisporum* as a separate species (pp. 889–902). Amongst the powdery mildews, molecular phylogenetic studies using sequences from three rDNA regions show that *Uncinula forestalis* is ancestral and the divergence may have occurred in the late Cretaceous; the new generic name *Caespitotheca* is introduced for the species (pp. 903–911). In addition, molecular phylogenetic studies on *Sawadaea* species placed them in seven clades, correlated with different sections of the host genus *Acer* and two geographical groupings; as the radiations in *Acer* occurred before those of *Sawadaea*, it is suggested that some of the species may have jumped onto maples from other hosts (pp. 912–922).

Septal ultrastructure is a key character in understanding phylogenetic relationships of basidiomycetes in particular. Electron microscopical studies of seven corticioid fungi showed all to have perforate parenthosomes, indicating that they do not belong to the cantharelloid or hymenochaetoid clades, and that an earlier report of imperforate parenthosomes in *Phanerochaete* was incorrect (pp. 923–926).

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The effects of grazing of a collembolan on *Hypholoma fasciculare* have been examined experimentally on wood blocks; coverage was drastically reduced and fewer mycelial cords were formed (pp. 927–935). A collembolan of Tertiary age entombed in Baltic amber has been found to be heavily infected by an *Aspergillus* species which is described as new (pp. 956–960). Macromycetes have been analyzed for their contents of numerous metals, but gold is rarely considered; an analysis of material of 89 species of ectomycorrhizal and saprobic fungi found the highest values in each

category in *Boletus edulis* and *Morchella esculenta*, respectively (pp. 951–955). A method to facilitate the cultivation of *Cordyceps unilateralis*, which produces anti-malarial red naphthoquinones, has been developed (pp. 936–940). In the entomopathogen *Entomophaga maimaiga*, which is specific to gypsy moth, AFLP analyses show the original Asian populations to be the most polymorphic, with few polymorphic loci occurring in North America – a result consistent with the species being introduced there (pp. 941–950).

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### THE CORONAMYCIN PRODUCER: A CASE OF MISTAKEN IDENTITY?

A mistake is revealed in the paper of Ezra *et al.* (2004) where an obvious *Penicillium* is called a *Streptomyces*. It is difficult to believe that this could be the case, and it gives me no pleasure in it being highlighted. However, ‘for the sake of science’ it is important that it is. This is clearly of relevance where secondary metabolites of huge potential significance are involved such as the so-called coronamycins. In addition, these reports influence which organisms are investigated for natural product discovery. A number of important additional issues are raised. For example, the crucial role of taxonomic specialists to natural product discovery, and over-reliance on PCR methodology.

I have discussed this case with other experts from both sides of the mycological and bacteriological fence, and we all agree that the organism is: (1) not a *Streptomyces*; and (2) a *Penicillium*. However, close examination of the published images reveals some actinomycete-like bacterial filaments in the background. The culture was evidently mixed, so it is unclear if the coronamycins were produced by the fungus, the bacterium, both, or partly by one and then modified by the other. Obviously the cultures were not purified by the use of appropriate media. This case demonstrates the importance of having taxonomists check the purity of cultures, although the scale of this mistake is larger than usual. Strobel’s defence (Strobel, Ezra & Castillo 2004), based on Gram staining and nuclear observations, is not convincing as the results were ambiguous, and the stain was not devised for fungi in any case.

Cavaletti & Monciardini (2004) were also convinced that the organism in question is not what was claimed, and attention has been drawn to other biochemical methods that can be used to distinguish fungi from other organisms (Paterson 2005). Now that it is realized that Ezra *et al.*’s cultures may have contained ‘invisible’ bacterial contaminants, DNA from those may have been inadvertently amplified by PCR, so generating bogus PCR results with respect to the verticillate fungus present. The problem of contaminants and

misidentifications in fungal DNA sequence databanks is known, and has been the subject of recent debate elsewhere (Hawksworth 2004). The key to resolving uncertainties such as that of the coronamycin producing organism(s) is the deposit of voucher cultures in public fungal genetic resource collections so they can be checked by other researchers (Ryan & Smith 2004). Sadly, in this case cultures were only deposited in ‘Montana State University Mycological culture collection’, where Ezra *et al.* are based, and subcultures are not being made available because of the potential economic value of the strain. In response to my request for a culture, Gary Strobel (pers. comm.) commented: ‘Unfortunately, the p-25 organism is licensed and patented to a company in Calif [*sic!*] and I am not at liberty to freely send the isolate to anyone other than the company’. More specific identification of the two organisms present is therefore impossible at this time.

Perhaps journals will need to specify the public collections in which vouchers must have been deposited prior to papers being accepted for publication to resolve this pernicious problem; further, researchers must decide whether they want to place their work in the public domain through publication in refereed journals, or make them available on an exclusive basis to a particular company for commercial exploitation; they cannot have it both ways.

Cavaletti, L. & Monciardini, P. (2004) Congruence between strain morphology and the 16S rRNA gene sequence. *Microbiology* **150**: 3093–3094.

Ezra, D., Castillo, U. F., Strobel, G. A., Hess, W. M., Porter, H., Jensen, J. B., Condron, M. A. M., Teplow, D. B., Sears, J., Maranta, M., Hunter, M., Weber, B. & Yaver, D. (2004) Coronamycins, peptide antibiotics produced by a verticillate *Streptomyces* sp. (MSU-2110) endophytic on *Monstera* sp. *Microbiology* **150**: 785–793.

Hawksworth, D. L. (2004) ‘Misidentifications’ in fungal DNA sequence databanks. *New Phytologist* **161**: 13–15.

Paterson, R. R. M. (2005) Fungus or bacterium and *vice versa*? *Microbiology* **151**: 641.

- Ryan, M. J. & Smith, D. (2004) Fungal genetic resource centres and the genomic challenge. *Mycological Research* **108**: 1351–1362.
- Strobel, G. A., Ezra, D. & Castillo, U. (2004) A question concerning the identity of *Streptomyces* sp. MSU-2110. *Microbiology* **150**: 3094–3096.

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## LONG-RANGE SELF-ORGANIZATION IN *Dictyostelium*

Just how the separate amoeboid cells of the slime-mould *Dictyostelium discoideum* organize themselves into multicellular structures has long fascinated developmental biologists. It has been known that nutrient-deprived amoebae aggregate as a result of chemical attraction (chemotaxis). The chemotaxis is through waves of adenosine 3'5'-cyclic monophosphate (cyclic AMP, cAMP) relayed from cell to cell, cores of outwardly rotating spiral waves self-organizing. However, the control mechanism for the system was unknown. Now, using seven mutants in the cAMP/protein kinase A pathway, Sawai, Thomason & Cox (2005) have been able to shed new light on this fascinating phenomenon. The mutants exhibited periodic signalling, but did not develop long-range self-organizing territories, and numerous separate spiral cores or cells were produced; the development of the cores is well-illustrated photographically and in a supplementary video available on-line. The *regA* and *erkB* mutants displayed around two- and four-fold increases in the number of

singularities, respectively. The chemotactic waves arising from different sources 'collide and annihilate each other before they can propagate very far', so preventing normal self-organization.

This study is claimed to be the first amongst living organisms demonstrating at the genetic level spontaneous symmetry-breaking in a self-organizing system. It is speculated that similar autoregulatory systems may operate in systems as disparate as heart muscle to prevent deleterious occurrences of spiral waves. Another example of the potential value of fungi as model systems for investigating fundamental biological processes, as stressed in *Mycological Research* last year (Gow 2004).

- Gow, N. A. R. (2004) New angles in mycology: studies in directional growth and directional motility. *Mycological Research* **108**: 5–13.
- Sawai, S., Thomason, P. A. & Cox, E. C. (2005) An autoregulatory circuit for long-range self-organization in *Dictyostelium* cell populations. *Nature* **433**: 323–326.

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## RESISTANCE VARIATION IN NATURAL PATHOSYSTEMS

Natural host-pathogen systems have received scant attention from plant pathologists and other mycologists, the pioneering work of Jeremy J. Burdon and co-workers being a notable exception (e.g. Burdon 1987, Thrall & Burdon 2000). Now a study has been carried out on the *Plantago lanceolata* – *Podosphaera plantaginis* pathosystem in south-west Finland where the host populations are isolated and only a small proportion of the populations is generally infected by the powdery mildew at any one time (Laine 2004). During the three-year study, the fungus infected only 2–5% of the populations, and some were infected one year and not the next, with new infections also occurring. Inoculation experiments were conducted using four isolates of the fungus and revealed 16 resistant phenotypes amongst 64 plants tested. Some populations belong to a single resistance type, while in others only

half showed identical resistance responses. No association between resistant populations and geographical location was found. The author suggests that the high levels of resistance in the host populations retards the spread of the fungus and so decreases the possibility of regional epidemics. It will be interesting to see whether this proves to be general model for natural pathosystems where fungus and host have co-existed for extended periods of time.

- Burdon, J. J. (1987) *Diseases and Plant Population Biology*. Cambridge University Press, Cambridge, UK.
- Laine, A.-L. (2004) Resistance variation within and among host populations in a plant-pathogen metapopulation: implications for regional pathogen dynamics. *Journal of Ecology* **92**: 990–1000.
- Thrall, P. H. & Burdon, J. J. (2000) Effect of resistance variation in a natural plant host-pathogen metapopulation on disease dynamics. *Plant Pathology* **49**: 767–773.